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Technical Report

Uptake of heavy metals from contaminated soils by Salt-Marsh plants

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Abstract

Spartina anglica, Puccinellia maritima and Aster tripolium, three common salt-marsh species in Western Europe, were grown in contaminated sediment from the port area of Antwerp (Belgium). Growth and levels of heavy metal contamination were compared with those of Spartina alterniflora, a common salt-marsh species from the United States. The plants were grown under waterlogged and drained soil conditions. In both cases was a high and a low soil salinity maintained.

The levels of heavy metals in the shoots of the plants were generally higher under drained conditions. The difference in salinity gave no obvious differences in metal levels in the shoots. Plants grown in the same sediment, after having allowed it to dry out and get aerated, had higher levels of heavy metals in their shoots.

The different plant species showed all different levels of metals in the shoots when grown under the same circumstances: P. maritima had the lowest levels, A. tripolium the highest. In A. tripolium there was also a significant difference in metal-levels in leaf- and stem material.

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Preface

This study was conducted at the Delta Institute for Hydrobiological Research, Yerseke, The Netherlands. It started in March 1983 with Dr. A.H.L. Huiskes, dept. of Experimental Botany, and Mr. J. Nieuwenhuize, dept. of Soil Chemistry, as chief investigators.

The study is one of a number of investigations and monitoring programs of the Delta Institute for Hydrobiological Research on the dynamics of anthropogenic substances in the environment. It also links up with the experiments performed at the US Army Corps of Engineers Waterways Experiment Station, Vicksburg, USA, to develop a bioassay procedure to assess the level of contamination of dredged sediments.

The project was financed by the U.S. Army Corps of Engineers Field

Verification Program, Mr. Charles C. Calhoun, Program manager. At various

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- Mr. J. van Soelen en Mr. M.M. Markusse for their assistance during the growth and the harvesting of the experiment.

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- Mr. L.J. Goud and Mr. J.C. Ruissen for taking care of the administration of the project.
- Drs. W.G. Beeftink and E.K. Duursma for reviewing the text and providing valuable additions and revisions.
- Mrs. M.J. van Leerdam-de Dreu, Mr. J.A. van den Ende and Mr. A.A. Bolsius for typing and for processing the text and the figures.
- Our families for allowing us to make regular journeys to the United States, for entertaining our foreign colleagues working in the same program.

1. Introduction

The boundaries between land and water have always been areas of major human activities. Large industrial complexes connected with harbour facilities are found everywhere in the world just in the interface between water and land as well as between sea and river. To keep the harbour basins open to shipping movements large quantities of sediments have to be removed regularly. These sediments are often polluted with anthropogenic substances (heavy metals, organic compounds, pesticides and oil residues) deliberately or inadvertently discharged into the water and partly adsorbed to the sediment particles. Depending on the levels of these pollutants the sediments may be a health hazard when used to create arable land or natural environments. In the United States of America one of the disposal alternatives is the creation of artifical marshes. Depending on the salinity of the dredged sediments and the disposal site these marshes may be fresh, brackish or saline.

The U.S. Army Corps of Engineers Waterways Experiment Station, Vicksburg, Mississippi is developing a bloassay for testing the quality of the dredged material, which in turn will provide evidence for the decision in what way the sediment will be disposed of.

In this study the procedure for the plant bicassay has been applied in experiments with some Dutch salt-marsh plant species growing on brackish contaminated sediment and using the North-American salt-marsh species Spartina alterniflora Loisel as a reference.

The specific objectives of the present study were:

- a. To evaluate the usefulness of the Waterways Experiment Station plant bioassay procedure by using other species and sediments;
- b. To investigate heavy metal uptake by a number of salt marsh species;
- c. To compare the heavy metal uptake of the North-American Spartina

 alterniflora Loisel with the uptake of native salt-marsh species, viz.

 Puccinellia maritima (Hudson) Parl., Aster tripolium L. and Spartina

 anglica Hubbard.

2. Exprimental design

The design of the glasshouse experiment was according to the bioassay procedure as has been described by Folsom et al. (1981a, b).

Figure 1 shows a diagram of the experimental unit used in the experiments. A small inner bucket rested on polyvinyl chloride (PVC) pipe inside a larger outer bucket. Six 6.35 mm diameter holes were drilled in the bottom of the inner bucket. These holes were covered with a 2.54 cm thick polyurethane sponge overlaid with a 2.54 cm (approximately) layer of washed quartz sand. The sand and the sponge acted as a filter to keep the

sediment/soil from draining out the bottom of the small bucket. The holes in the small bucket allowed water movement into and out of the sediment. The water level in the inner buckets was maintained by filling the space between the outer and inner bucket up to a certain height with water.

2.1. Sediment preparation

The polluted sediment used in this study was dredged by one of the institute's research vessels R.V. "Jan Verwey" using a "Van Veen" grab. The dredging place was just outside the big shipping lock "Zandvlietsluis" in the Belgian part of the Westerschelde estuary (Fig. 2). This sediment is rather homogenous in texture and concentrations of various substances, as it is well mixed by tugs towing iron bars across the entrance of the lock in attempt to resuspend the sediment and a subsequent removal of it with the outgoing tides.

Immediately after dredging 8 samples of the sediment were taken randomly, to analyse the sediment for the various heavy metals. The dredged sediment, transported in thoroughly cleaned plastic containers, was spread out on large flats lined with PVC sheets to let it dry out. Excess water was siphoned off. After about three months, the water content of the sediment was 60.1 % on weight basis.

2.2. Plant material

The experiment was divided in two parts for reasons of available space in the greenhouse. The first year an experiment was done with <u>Spartina alter-</u>

niflora and Spartina anglica. Spartina alterniflora was obtained from WES transported by air. The plants were commercially grown from seeds and had 4 to 5 shoots, approximately 20 centimeters high. Spartina anglica is at present the most abundant Spartina species in the Netherlands. It was imported by the Public Works Department from Great Britain in 1924 and 1925 for land reclamation purposes and has spread since then naturally or by deliberate planting to almost all salt marshes in the Netherlands. For the experiment cuttings with 4 to 5 shoots were taken in the field and transplanted to flats filled with potting compost to allow them to root. The second year an experiment was done with S. alterniflora-cuttings and cuttings of Aster tripolium and Puccinellia maritima. The inner buckets were filled with a known amount of sediment (7000 g approximately). In the sediment the rooted cuttings were planted (or in the first experiment seedlings of S. alterniflora). The water and salinity regime in the buckets was: a the outer bucket filled with artifical seawater to a height just above the surface of the sediment in the inner bucket to maintain a waterlogged situation.

- <u>b</u> the outer bucket filled with artificial seawater of 50% of the normal strength, to a height just above the surface of the sediment in the inner bucket.
- c the outer bucket filled to a height of 5 cm with artifical seawater of normal strength, to keep a certain soil moisture by capillary rise.
- d the outer bucket filled to a height of 5 cm with artifical seawater of 50% of the normal strength.

Each treatment had 5 replicates for each species. As a reference <u>S. alterniflora</u> was also planted in buckets filled with a reference soil, obtained from WES. This is a loamy soil, which was mixed with a fertilizer by WES.

In the first experiment four buckets were kept waterlogged and four buckets were kept under dry conditions with artifical seawater of 50% of the normal strength. In the second experiment only two buckets with WES soil were kept as a reference under dry conditions (i.e. the outer bucket filled with 5 cm of artificial seawater of 50% of the normal strength.

In the waterlogged situation the outer buckets were regularly adjusted to the level fixed at the start of the experiment with artificial seawater. In the other buckets the level of 5 cm was kept constant too but the plants in these buckets were daily watered with demineralized water from the top.

Once a week the watering was performed with artificial seawater.

2.3. Harvesting of the plants

Ninety days after the start of the experiment the shoots of the plants were cut at the soil surface with a plastic knife to avoid metal contamination. All handling of plants and sediments was done wearing plastic gloves. Almost all plants of <u>S. anglica</u> flowered after ninety days. The plants growing in the "upland" situation in the first experiment were harvested four weeks later, as the salinity levels in these containers were established four weeks after planting. Per treatment were for each species also one root sample and one soil sample taken from a bucket. This was only done in the first year as the buckets in the second year were kept intact for a regrowth experiment.

The shoots and roots were thoroughly rinsed with demineralized water and blotted dry with filter paper. Fresh weight of whole shoots was

measured for <u>S. alterniflora</u>, <u>S. anglica</u> and <u>P. maritima</u>. The shoots of <u>A. tripolium</u> were divided into stem, leaves and - if present - inflorescences. These fractions were separately weighted and subsequently analysed. For <u>S. alterniflora</u> and <u>S. anglica</u> also the number of tillers and the total length of the shoots were measured.

2.4. Analysis of various components in plant samples

After the measurement of the fresh weight the samples were kept at -20° C (-4° F) awaiting analyses. For the analyses the samples were freeze-dried to constant weight. The dried samples - both plant and sediment symples - were then ground in a agate mill to avoid metal contamination. All handling of samples was done, using plastic gloves. The plant samples were subsequently digested with HNO_3 conc. and H_2O_2 (30%). The destruate was brought to a volume of 50 ml by adding demineralized water. This volume contained still 2 ml HNO_3 . All elements were measured in this watery extract. Ca, Mg, Fe, Mn and Zn were measured by means of atomic absorption spectrometry, using an air-acetylene flame. Na and K were measured by means of flame - emission spectroscopy using also an air - acetylene flame.

Pb, Cd and Cu were measured on the atomic absorption spectro photometer in combination with a graphite furnace. Pb and Cd were measured by means of the standard addition method.

As was determined by hydride generation and atomic absorption spectroscopy.

All measurements were performed with a Perkin Elmer model 2380 instrument.

2.5. Analysis of various components in sediment samples

The sediment samples were freeze-dried to constant weight and ground in an agate mill. The samples were sieved through a 2 mm sieve. Due care was taken to avoid heavy-metal contamination during the treatment of the samples. The chloride content of the samples was measured by potentiometric titration with AgNO₃ in a watery extract. CaCO₃ was measured by shaking 3 g of dry soil with 15 ml demineralized water and 7 ml HCl (25%). The produced CO₂ was measured volumetricly.

The particulate organic matter content was measured by pyrolysis of the sample and weighing of the produced ${\rm CO}_2$, adsorbed at an adsorption complex. ${\rm P}_2{\rm O}_5$ was determined colorimetrically by the vanadate method (Andersen, 1975). Total nitrogen was measured as ${\rm N}_2$ after pyrolysis in a Carlo Erba model 1400 nitrogen analyzer. The pH was measured as pH KCl. All metal-ions were determined as total contents after destruction of the sample with a HCl-HNO $_3$ mixture (3:1) and a subsequent destruction with ${\rm H}_2{\rm O}_2$ (Schramel et al., 1982). All ions were analysed in the destruate with the Atomic Absorption Spectrophotometer in combination with a graphite furnace.

3. Results

3.1. Sediment analyses

Immediately after dredging 8 soil samples were taken from the total mass of 1000 litres of sediment dredged near Antwerp (Fig. 1). The results are given in Table 1. (The individual results are given in Appendix A). Also in

Table 1 are shown the results of soil samples taken after the experiment with S. alterniflora and S. anglica in 1983. As these figures are based on just one sample no statistical treatment of the data could be performed and they have to be treated as an indication.

The figures for the moisture content of the soil and the salinity under waterlogged and drained conditions and the high and low salinity levels may indicate that the envisaged treatments were not very well maintained. Under drained conditions the salinity was low at both salinity levels, while under waterlogged conditions the salinity was comparatively high at both levels. This means that with the interpretation of the results the fact has to be kept in mind that the treatments applied were waterlogged conditions coupled with high salinity and drained conditions coupled with low salinity. Although the results of the analyses of the plants will be presented as results for the four treatments, in the discussion the data will be related to the above mentioned findings.

3.2. Plant analyses

As there are at certain points significant differences between the data of Spartina alterniflora plants grown in 1983 and plants grown in 1984, the results of the two parts of the experiment are presented separately with S. alterniflora s a reference.

3.2.1. Growth parameters

Figure 3 shows the fleshweight of the shoots for S. alterniflora in the experiments of 1983 and 1984, both on dredged sediment and on WES soil.

There is a difference in average fresh weight per plant between plants grown on WES soil and plants grown on dredged sediment, this could be due to a difference in soil fertility. No significant difference could be found between the various treatments and the two years (analysis of variance). The results obtained from plants grown or WES-soil were not statistically treated, as they were too few and too different from the results on the dredged material. The inidividual results are given in appendix B. Fig. 4 shows the average amount of tillers produced per plant. There is a significant difference between the average number of tillers produced in 1983 and in 1984 and also a significant difference in 1983 between drained and waterlogged circumstances. The difference in tiller production could be due to the fact that in 1983 the plants were grown from seedlings and in 1984 from cuttings.

In fig. 4 the number of tillers of <u>S. anglica</u>, the European <u>Spartina</u>, is also shown, together with the LSD (= least significant difference) of the findings of both <u>Spartina</u>-species in 1983. The difference between the two species is obvious together with the difference between the number of tillers under drained and under waterlogged conditions. All plants of <u>S.</u> anglica were flowering at the end of the experiment.

In fig. 5 the average fresh weight of the two <u>Spartina</u> species grown in 1983 is compared. An analysis of variance showed a significant difference between the two species, but no significant difference was found due to the four treatments. Appendix C gives the individual results.

Fig. 6 shows the average fresh weights of the part of the experiment performed in 1984. The species studied were <u>S. alterniflora</u>, <u>P. maritima</u> and <u>A. tripolium</u>. Comparison between the results is questionable as the species are not as closely related as the Spartina's are (same genus).

Moreover A. tripolium is a forb with a completely different growth form as compared with the other two species, which are grasses. The latter two species produce a negligible amount of stem material as compared with A. tripolium. For A. tripolium stem material and leaf material are processed separately both chemically and statistically. No significant difference between the results from the various treatments could be found. There is a significant afference between the species however. This is not surprising as the stem fraction was treated statistically as a separate species. The weights of this fraction though were substantially lower as compared with the leaves fraction or the shoots of the grasses. The results of Aster tripolium show a clear difference in behaviour under the various treatments. Under low salinity conditons the plants flowered, which did not occur under high salinity conditons. Under high salinity conditons the plants form a substantial amount of dead leaves, while the amount of fresh leaves do no differ significantly. The turnover rate of leaves must therefore be higher under high salinity conditions. This has been shown for other halophytes as well (Waisel 1972). The difference between the results from high and low salinity treatments for Aster tripolium could indicate that in 1984 the two salinity levels might have been established properly, contrary to the results shown for 1983 (Table 1). This fact however could not be checked, as no soil samples were taken, for the reason that the buckets with their contents were saved for a regrowth experiment.

3.2.2. Plant analyses

Table II and Table IV show the average results of the analyses of the

shoot material of the experiments of 1983 and 1984 respectively. The results of the experiment performed in 1983 (Table II) are the means of samples from five replicate treatments. The results shown for <u>S. anglica</u> grown under high salinity and waterlogged conditions are the means of three samples, as two plants showed a strongly reduced growth as compared with all other plants.

In Table III the significance of the individual figures is shown.

Although Table III shows considerable overlaps in significant differences there are some general tendencies. The influence of inundation or drainage is present for Mm, Cu, As, Na, K, Ca and Mg and to a lesser extent for Zn. Mm, Na, Ca and Mg were takenup more under inundated circumstances, while Cu, As, K and to a certain extent Zn were taken up in higher quantities under drained conditions.

Cd showed a tendency to be taken up more by <u>S. alterniflora</u> while Fe seemed to be taken up in higher amounts by <u>S. anglica</u>. The pattern for Pb is not clear. The difference in high and low salinity treaments is unclear as was already suggested in paragraph 5.1.

Table IV shows the results of the experiment performed in 1984 S. alterniflora was mainly used in this experiment to link it up with the findings in 1983.

Table V gives the significant differences between the various results. The table presents the significant differences between the species — whereby the stem — samples of <u>A. tripolium</u> are treated as a separate species — and between the treatments, which gives a better survey in this case as compared with the method used in Table III. For most metal — levels the leaves of <u>A. triplium</u> score highest in a number of cases are these differences significant. The grasses have significantly lower levels of Cd in their

shoots as compared with the forb A. tripolium. This is also the case for Cu, Zn and to a lesser extent for Fe, Zn and Na. As-, K- and Ca levels show significant differences for all species. Mn- and Mg- levels are significantly lower in the stems of A. tripolium, and in P. maritima-shoots, while Pb seems to be accumulated especially in the stem of A. tripolium.

No significant differences due to the treatments were found in levels of Pb, Fe, Man and Mg. Cd, Cu and Zn were taken up more under drained circumstances. The level sof As differed significantly between all treatments. Na-, K- and Cu-levels showed no obvious difference.

An analysis of variance performed on the results of <u>S. alterniflora</u> of 1983 and 1984 showed significant differences between the two experiments for levels of Fe, Mn, Cu, As, Na, K and Ca. The shoots of <u>S. alterniflora</u> had in 1984 significantly higher levels of Fe, Mn, Na, K and Ca. The levels of Cu and As were lower in 1984.

4. Conclusions and recommendations

4.1. Conclusions

The division of the total experiment into two parts, apread over two consecutive years may have obscured some of the differences in plant uptake of heavy metals and in the influence of the four treatments. Firstly the plant material of <u>Spartina alterniflora</u> was different in the two years. In 1983 young plants, grown from seedlings were used, in 1984 the plants were grown from cuttings taken from older plants. This may explain the difference in tiller production in 1983 and 1984. In 1983 there was a significant difference in tillers production under drained and waterlogged conditions (Fig.

4). In 1984 however no significant difference was found and the mean number of tillers was generally higher. This was also the case with the plants grown on the reference soil obtained from the Waterways Experiment Station.

Another difference between the experiment of 1983 and that of 1984 was that in 1983 sediment was used that had been dredged only two and a half months before the start of the experiment, while in 1984 the same sediment was used which by then had had about fifteen months to settle and ripen. The sediment, even over these fifteen months, never dried out; at the start of the experiment in 1984 it was still a "mousse".

These features were the reason why the experiments of the two years were discussed separately. An indication for the difference in soil conditions in 1983 and 1984 might be the higher levels of especially Mn and Fe in the shoots of <u>S. alterniflora</u>. It is known that a lower redox potential may result in a higher availability of these metal ions (Rozema and Roosenstein, 1985).

Analyses of salinity in 1983 showed that the high salinity - drained treatment had a lower salinity than the high salinity - waterloged treatment (Table VI). This was undoubtedly due to the watering of the pots with tap water to prevent serious desiccation of the top layer of soil in the pots. This watering apparently washed down the salt, although once a week the pots were watered with artifical sea water. Although in 1984 the drained pots were watered more often with artificial sea water, the results (Table V) show that the difference in the two treatments was still not clear. As the pots were used for a sequential regrowth experiment, no soil samples were taken. This resulted effectively in three treatments: high salinity - waterlogged, low salinity - waterlogged and low salinity - drained.

The differences in heavy-metal levels in the different plant species is apparent especially in the experiment of 1984. For a bioassay procedure it is imperative to make a choice for one single-standard species. This may be Spartina alterniflora in the American situation but in the Dutch situation with in general a greater species diversity in the salt marsh it may be difficult to make a choice. Spartina anglica is a dominant species in the Dutch salt marsh and could therefore qualify. Puccinellia maritima is known as a low accumulator, while Aster tripolium is a high accumulator of heavy metals (Beeftink et al., 1982). The last species is eaten as a vegetable in the Netherlands and is in that quality subject to the standards of heavy metal contamination of the Ministry of Agriculture. It is tempting to use these standards for the bioassay with A. tripolium, to decide whether a soil is contaminated or not, but there is no jurisdiction with respect to this matter.

4.2. Recommendations

- The bioassay procedure, developed at Waterways Experiment Station, provides good method to assess the contamination level of a certain amount of dredged material.
- It has to be decided however, which species should be used as a European test organism, as different species accumulate heavy metals at different levels.
- It is clear from the results over the two years that the time between dredging and bioassay procedure is decisive for the availability of the various metal ions. It is therefore imperative to take this time into account when the results of a bioassay procedure are analysed.

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- Figure 1. Diagram of the experimental unit, used in the experiments.
- Figure 2. Map of the south-west Netherlands, with the position of the dredging area.
- Figure 3. Fresh weight of shoots of <u>Spartina alterriflora</u> grown in 1983 and 1984 under different treatments.
- Figure 4. Number of tillers of <u>Spartina alterniflora</u> and <u>Spartina anglica</u> grown in 1983 and 1934 under different treatments.
- Figure 5. Fresh weight of shoots of <u>Spartina alterniflora</u> and <u>Spartina</u>

 anglica grown under different treatments.
- Figure 6. Fresh weight of shoots of <u>Spartina alterniflora</u>, <u>Puccinellia</u>

 maritima and <u>Aster tripolium</u> grown under different treatments.

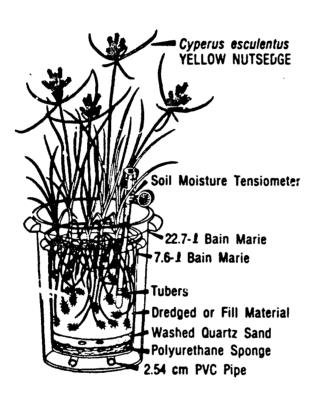
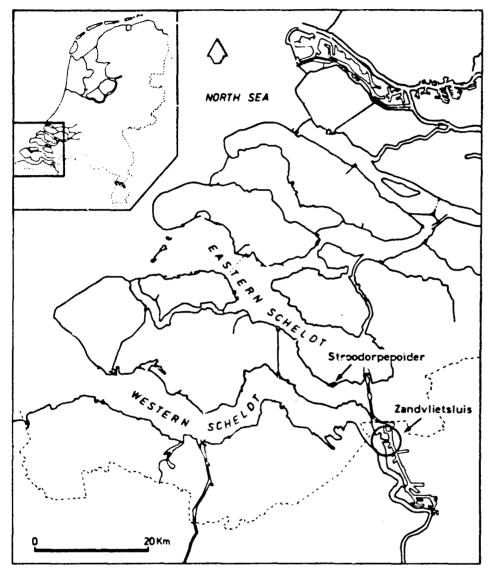
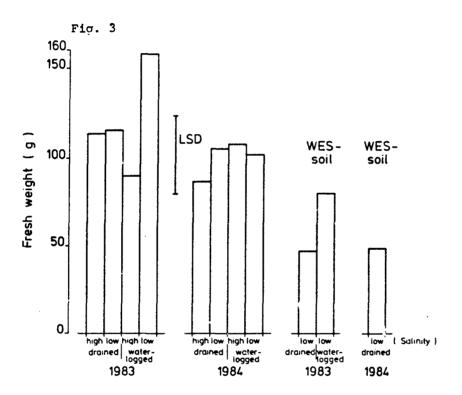


Fig. 2





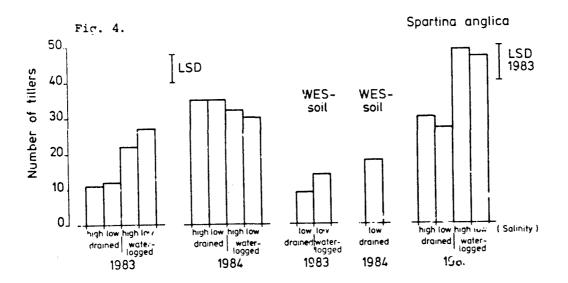
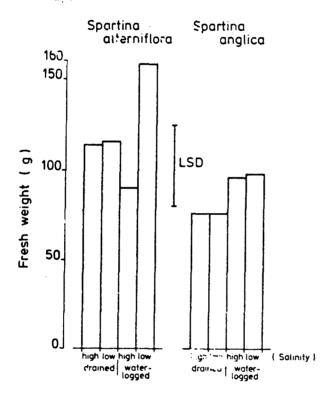
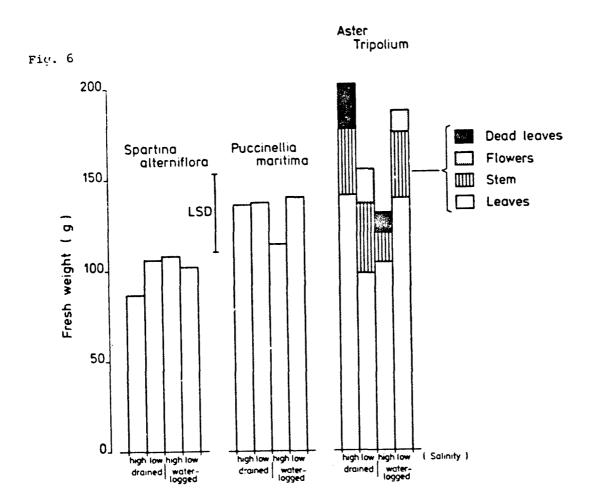


Fig. 5





le 1. Analysis results of soil samples taken immediately from the dradged sadiment and taken after the sadiment was left tot dry out en plants were grown in it for ninety days.

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reine emplica salinity erloged	14.9			87.2	2.13	27.92	119	1.1	4.7	0.29	9.94		11.0	140	3.80	*	17.	
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rtine alterniflora h selimity	13.0			76.8	0.43	5.62	611	7.6	3.6	0.27	43.4		7.11	138	3.65		794	
rtina alterniflora aslinity erlegad	•••			6.6	2.12	21.29	126	7.6	*:	0.29	46.6		11.3	142	3.90	.	\$	
gries alterniflors selfaity Amed	13.6			62.0	0.21	3.39	128	7.5	3.5	0.26	36.3		10.7	118	3.59	z.	*	
reference soil																		
ref. soil grins elterniflors ; selimity arlogged	0.65			36.5	0.0	1.15	22	•	6.0	0.07	26.8		÷:	112	1.23	•	37	
ref. soil grins elterafflors selimity	0.47			30.9	0.0	2.18	21	4.6	0.	0.05	23.6		1.6	104	1.51	•	2	
•																		

Table II. Amounts of metal lons (given in mg.kg on dry weight basis) in shoots of Spartina anglica and Spartina alterniflora grown under different salinities and soil moisture conditions. The levels of Na, K, Cd and Mg are given in mg.g.

		Spartina	anglica		Spa	Spartina alt	alterniflora		
	high salinity	inity	low salinity	Inity	high salinity	linity	low salfafty	nity	L.S.D.
	1 undated drained	drained	fuundated	drained	inundated drained	drained	Inundated	drained	
3	0.130	0.118	0.112	0.074	0.214	0.284	0.170	0.228	0.144
P	0.522	0.474	0.286	0.482	0.388	0.490	0.368	0.510	0.176
Pe	74.2	62.8	62.6	8.69	8.65	52.0	8.65	9.99	11.5
£	117.0	92.6	104.0	80.0	116.2	8.06	100.2	82.6	24.6
3	3.30	4.00	3.50	4.96	3.44	5.38	2.58	5.30	1.37
uZ	14.2	20.6	17.6	26.4	23.2	36.4	16.6	26.4	11.2
As	0.162	0.282	0.158	0.258	0.152	0.306	0.154	0.266	0.077
2	33.88	23.04	31.46	20.66	24.94	16.06	22.28	14.60	3.71
×	8.52	13.86	8.92	13.62	12.62	15.98	10.18	15.20	3.05
đ	3.10	2.32	2.78	2.08	2.84	2.54	2.70	2.36	0.52
¥.	3.70	1.94	3.54	1.82	2.18	1.86	2.34	1.72	0.51

Table III. Least significant differences (LSD) in heavy-metal concentrations in shoots of <u>Spartina anglica</u> and <u>Spartina alterniflora</u>.

The figures 1 to 8 represent the various treatments:

Spartina anglica	high salinity	inundated	1
		drained	2
	low salinity	inundated	3
	-	drained	4
Spartina alterniflora	high salinity	inundated	5
		drained	6
	low salinity	inundated	7
		drained	8

The underlined figures differ not significantly of each other.

Cd	4_	3	2	1	7	_5	8	6
РЪ	3	7		2	4	6	8	1
Fe	6	8	5	7	3	2	4	1
Mn	4	8	6	2	7:	3	5	1
Cu	7	1	5	3	2	4	8	6
Zn	1	7			5		8	6
As	5	7					2	6
Na	8	_6	4	7	2	5	3	1
K	1_	3	<u>-</u>	5	4	2	8	6
Ca	4	2	8	6	7	3	5	1
Mg	8_	4	6	2	5	7	3	1

Table IV. Amounts of metal ions (given in mg.kg. on dry weight basis) in shoots of Sparting alterniflors, Aster tripolium (leaves and stem) and Puccinellia maritima grown under lifferent salinities and soil moisture conditions. The levels of Na, K, Ca and Mg are given in mg.g.

		Sparting	Sparting alterniflors	or.	Puccin	nellia maritima	ritime		Aste	r tripoliu	Aster tripolium (leaves)		Aste	Aster tripolium (stem)	in (eten)	
	bieb e	bish seliater	low salinity	Ifalty	high sel	falty	high salinity low salinity	nity	high sal	high salinity	low selinity	ity	high se	high salinity	low salinity	nity
	Inumdated	drained	inumdated drained inumdated drained	dreined	12	•	Irained inundated drained	drained	inundated	drained	inundated drained inundated drained	drained	inundated drained inundated drained	drained	Inundated	drained
2	0.186	0.362	0.156	0.214	0.114	0.194	0.114	0.300	0.222	0.580	909.0		0.260	0.566	0.228	₩06.0
£	0.380	0.368	0.212	0.432	0.712	0.504	0.384	1.056	0.520	0.640	0.808	0.568	1.354	0.996	0.500	0.468
	253.0	153.6	176.2	160.8	75.4	92.2	143.6	79.8	259.6	208.2	168.0			405.8	185.8	85.2
	126.6	100.0	16.0	115.8	9.69	68.2	4.19	71.4	61.0	130.8	141.0			40.2	29.5	30.8
. 3	3.40	3.08	3.16	3.42	4.22	5.18	3.10	3.64	5.18	7.60	5.38	6.40	13.24	19.00	10.18	10.88
. 5	₹0.4	21.4	20.0	23.0	11.4	13.4	10.0	13.6	29.0	37.0	45.4		23.4	34.0	20.6	25.0
1	0.200	0.144	0.122	0.194	0.230	0.228	091.0	0.354	960.0	0.030	0.112	0.070	0.298	0.242	0.136	0.118
4	24.62	19.8	22.24	19.44	14.08	9 .0 9	8.74	49.9	90.09	57.42	69.14	48.72	10.14	19.36	20.28	16.94
.	14.16	25.12	12.32	11.98	27.84	18.86	15.46	19.90	28.84	29.68	22.54	29.36	6.12	10.98	7.98	11.68
3	3.28	3.58	3.36	3.48	1.58	1.14	1.22	1.36	4.92	5.18	8.48	7.60	2.02	2.46	3.12	2.74
*	1.24	1.74	2.08	2.18	1.84	0.93	1.28	1.10	2.12	2.12	2.32	2.14	1.70	2.06	1.26	1.26

Table V. Least significant differences (LSD) in heavy-metal concentrations.

The figures ' co 4 respresent the plant species. The letters A to D represent the treatments Spartina alterniflora = 1; Puccinellia maritima = 2; Aster tripolium-leaves = 3; Aster tripolium-stems = 4. High salinity, inundated = C, High salinity, drained = A, Low salinity inundated = D, Low salinity, drained = B. The underlined symbols differ not significantly from each other.

		Speci	.es		T	reats	ents	
Cd	2	1	4	3	<u>c</u>	D	<u>A</u>	В
РЪ	1	3	2	4	D	A	В	<u> </u>
Fe	3	_1	2	4	D	В	<u>A</u>	<u>c</u>
Mn	4	2	1	3	<u>c</u>	<u> </u>	D	<u>B</u>
Cu	1	2	3	4	D	В	С	A
Zn	2	1	4	3	C	D		В
•								
As	3	1	4	2	Þ	A	В	С
Na	2	1	4	3	В	A	<u> </u>	D
K	4	1	2	3	D	<u>B</u>	c	A
Ca	2	4	1	3	<u>c</u>	<u> </u>	В	D
Mg	2	4	1	3	В	<u> </u>	D	<u> </u>

8. Appendices

Appendix A - Complete results of the experiments

- A. Analyses of the eight soil samples taken randomly, immediately after dredging.
- B. Heavy metal concentrations and yield of Spartina alterniflora (1983).
- C. Heavy metal concentrations and yield of Spartina anglica (1983).
- D. Reavy metal concentrations and yield of Spartina alterniflora (1984).
- E. Reavy metal concentrations and yield of Puccinellia maritima (1984).
- F. Heavy metal concentrations and yield of Aster tripolium leaves (1984).
- G. Heavy metal concentrations and yield of Aster tripolium stems (1984).
- H. Quality control analysis-methods.

Appendix A. Analysis results of 8 soil samples, chosen at random, from 1000 litres contaminated material, samples with the "van Veen" grab near the Zandvlietsluis (Antwerp) - 6th January 1983. Calculated on dry basis except if mentioned

			•	moisture	NaC1	$(100 \text{ g})^{-1}$		•			
(meq (100 g) ⁻¹	zeq (100 g) ⁻¹	(100 g dry) ⁻¹	dry	g NaCl 1	8	(100 g) ⁻¹	n 6	ĸ ģ	××
Lebar.	င် (၁၅)	g X	4	<	٩	ر		5	ā.	3	:
27096	14.1	30.4	2,35	174.4	1.73	9.91	133	2	6.9	4.3	0.318
27097	14.5	32.3	2,31	173.4	1.69	9.77	13(0	7.1	4.5	0.323
27098	13.2	29.7	2.19	174.8	1.59	9.13	12	9	7.0	4.7	0.321
27099	13.7	29.9	2.15	174.1	1.38	7.91	132	2	7.0	5.4	0.323
2/100	14.6	33.2	2.26	175.5	1.64	9.35	12	•	7.1	9.4	0.318
27101	15.8	34.0	2.23	173.8	1.52	8.76	12	\$	7.0	5.5	0.321
27102	14.3	30.1	2.15	171.8	1.64	9.55	13	9	7.0	6.4	0.329
27103	13.2	29.5	2.17	171.7	1.66	89.6	13	2	7.1	5.1	0.324
	14.2	31.3	2.23	173.7	1.61	9.26	12	σ	7.0	4.9	0.322
S.D.	3.8	1.8	90.0	1.4	0.11	99.0		4	0.1	9.4	0.004
	mofeture		u dd	Z mdd	.	mdd	mdd	mdd	C/N	ratio	-
	(100 g wet)	-1 clay	•								
Labur.	×	•	3	Pb Fe	a ,	చె	Zu	ප්			
27096	63.6	44.9	10.1		.95	86	498	197	13	٠.5	
27097	63.4	39.9	10.6		.46	92	510	100	13	6.	
27098	63.6	40.0	10.7		.76	95	867	203	14	9.	
27099	63.5	38.3	10.7		90'	93	867	200	16	.,	
27100	63.7	40.5	10.8	140 3.	60'	88	519	201	14.	5.	
27101	63.5	41.2	10.8		.79	88	207	201	17		
27102	63.2	40.0	10.7		,46	89	510	203	14	٠ •	
27103	63.2	41.2	10.6		.13	06	504	204	15	.,	
Hean	63.5	40.8	10.6		2.96	06	206	201	15	- ;	
S.D.	0.2	1.9	0.2	3	.30	m	œ	7	-	ů,	

Appendix B. Heavy metal concentrations and yield of Spartina alterniflora (1983).

							; ;				mo. 0 - 1		
Pot nr.	Fresh weight	nr. of			-	<u>.</u>					0 0		
	shoot		8	Pb	Fe	Ŧ	ņ	Zu	As	Na	×	రో	ħ
drained,	high salinit	Infty											
5 6	118.77	10	0.37		48	113	5 1	94	0.27	17.4	16.0	2.9	1.7
27	121.40	∞	0.11		26	96	5.9	27	0.44	16.5	13.6	3.1	2.2
28	89.60	10	0.69		54	29	6.1	65	0.14	15.9	15.1	3.0	2.1
29	158.87	15	0.13	0.29	45	105	4.1	5 6	0.31	16.6	13.0	2.2	1.9
30	81.31	10	0.12	0.95	27	73	5.7	18	0.37	13.9	22.2	1.5	1.4
drained.	low salinf	ifty											
36	132.06	13	0.24		20	82	5.1	30	0.23	16.7	14.1	2.3	1.7
37	112.48	14	0.15		68	26	5.7	18	0.31	14.5	16.7	1.9	1.4
38	91.52	11	0.19	09.0	62	29	5.8	70	0.31	12.8	15.9	2.6	1.9
39	152.72	14	0.42		44	130	5.4	41	0.22	13.1	15.7	3,3	2.2
07	93.69	6	0.14		19	78	4.5	23	0.26	15.9	13.6	1.7	1.4
Inundated	1, high sa	ilinity											
21	85.50	15	0.17		9	74	4.6	3	0.21	21.6	10.7	2.7	2.7
22	131.91	22	0.32		29	133	5.2	27	0.18	24.5	13.0	2.9	2.0
23	55.84	10	0.17		61	153	2.9	22	0.13	33.8	13.2	3.1	3.0
24	139.86	33	0.29	0.43	55	122	2.4	18	0.14	24.7	12.7	2.7	1.6
25	38.99	29	0.12		52	66	2.1	19	0.10	20.1	13.5	2.8	1.6
Inundated	1, low sal	linity											
31	155.28	23	60.0		63	109	2.2	16	0.15	24.3	9.5	2.9	2.3
32	99.90	27	0.19		71	105	2.5	14	0.16	23.1	9.5	3.0	2.3
33	119.32	24	0.15	0.49	99	83	2.2	17	0.16	22.6	11.3	2.2	1.6
34	158.11	27	0.19	•	63	108	2.9	17	0.17	22.6	9.5	3.0	2.4
35	258.83	32	0.23	•	46	96	3.1	19	0.13	19.8	11.1	2.4	2.1

Appendix. C. Beavy metal concentrations and yield of Spartina anglica (1983).

					DOG -	1 00					1- 11- 110.8		
Pot nr.	Fresh weight	nr. of tillers	5	ă			Ë	\$	-	Ž))) b	_	\$
	1001		3	۲. م	ภ	E	3	117	Š	2	4	3	<u>5</u>
drained,	high sali	Inter											
•	73.66	25	0.09	0.59	77	98	4.9	74	0.30	24.5	13.4	1.9	1.7
7	87.38	24	0.29	0.46	26	122	2.4	19	0.15	27.6	16.0	2.9	2.2
∞	73.30	23	0.07	0.30	29	89	3.6	18	0.35	18.9	13.9	2.0	1.6
Φ.	87.91	53	0.03	95.0	47	8	5.6	12	0.32	23.2	7.8	2.7	2.4
21	59.34	24	0.11	0.56	6 7	9/	6.5	30	0.29	21.0	18.2	2.1	1.8
drained,	low salin	ity											
16	116.30	34	0.05	0.41	59	99	3.5	20	0.18	20.0	8.8	2.1	1.0
17	56.14	24	90.0	0.63	69	79	5.9	31	0.25	21.5	17.0	2.3	1.8
18	79.50	20	0.11	0.33	72	. 83	7.2	36	0.23	24.5	15.9	2.0	2.0
19	62.68	26	0.05	0.35	83	82	3.7	18	0.34	18.6	13.4	1.7	1.6
20	63.11	31	0.10	69.0	99	88	4.5	27	0.29	18.7	13.0	2.3	1.8
Inundated	d, high sa	linity											
-	18.14	18	0.29	1.4	229	16	4.9	32	0.30	39.8	10.2	4.2	4.8
7	3.12	7	0.23	1.2	244	64	8.9	21	ı	43.9	10.4	4.1	5.5
m	93.83	51	0.14	64.0	69	122	3.5	12	0.16	34.6	8.8	3.6	3.6
∢.	63.24	34	0.13	0.50	75	127	3.6	18	0.17	37.7	9.6	3.2	4.3
'n	131.82	62	0.12	0.58	79	102	2.8	13	0.16	29.3	7.4	3.1	3.2
inundate	d, low sal	inity											
11	127.17	57	0.11	0.21	46	112	3,3	14	0.14	33.1	8.7	3.0	3.5
12	105.82	58	0.04	0.21	53	06	2.8	15	0.11	31.7	7.7	2.6	3.6
13	120.06	47	0.14	0.40	74	98	3.0	17	0.19	28.7	7.9	2.7	4.0
14	46.19	27	0.16	0.26	7.5	122	4.6	5 6	0.22	32.7	11.1	2.8	3.4
15	89.96	46	0.11	0.35	65	86	3.8	16	0.13	31.1	9.2	2.8	3.2

Į

Appendix. D. Heavy metal concentrations and yield of Spartina alternifiora (1984).

											mo. o		
	Fresh	nr. of											
Pot nr.	weight shoot	tillers	ষ্ট	æ	Fe	Æ	3	Zu	As	N.	×	రో	₹
drained.	high salinity	Inity											
41	125.47	33	0.42	0.46	183	116	3.4	17	0.17	21.5	23.5	3.4	1.9
42	56.65	56	0.28	0.30	131	78	2.8	17	0.11	18.5	24.3	3.1	1.5
43	84.34	37	0.40	0.54	158	101	3.3	25	0.15	18.0	27.0	3.5	1.8
44	92.59	43	0.39	0.36	159	107	3.4	28	0.12	22.0	26.8	4.0	1.9
45	67.84	37	0.32	0.18	137	98	2.5	20	0.17	19.3	24.0	3.9	1.6
drained,	low salin	ifty											
46	69.66	37	0.35	0.42	189	124	3.6	28	0.22	17.3	11.9	3.9	2.0
47	107.30	40	0.22	0.36	137	116	4.2	23	0.17	22.4	12.7	4.3	2.5
48	84.40	37	0.35	0.54	161	76	3.0	25	0.20	17.0	14.6	3.2	1.6
69	100.90	31	0.12	0.64	226	101	3.0	23	0.25	22.3	12.6	2.9	2.2
20	136.12	32	0.03	0.20	16	144	3.3	16	0.13	18.2	8.1	3.1	5.6
inundated	d, high sa	ilinity											
51	, ,	39	0.19	0.16	104	124	3.7	22	0.11	26.5	22.0	3.7	2.8
52	126.88	44	0.26	0.20	172	114	3.1	23	0.12	20.8	10.8	2.8	2.0
53	91.47	22	0.17	0.30	203	139	3.3	21	0.14	24.0	13.0	3.1	2.0
54	94.24	23	0.12	0.38	282	122	3.4	18	0.26	27.8	13.5	3.4	2.2
55	119.73	32	0.19	0.86	504	134	3.5	18	0.37	24.0	11.5	3.4	2.2
Inundated	_	Infty											
56	58.29	21	0.11	0.40	128	106	3.6	77	0.12	26.3	15.6	3.9	2.5
57	112.41	26	0.11	0.20	365	103	3.0	16	0.12	21.4	9.7	3.3	2.1
58	124.77	39	0.20	0.24	167	143	2.9	19	0.11	21.0	13.1	3.5	1.9
59	95.57	36	0.13	0.10	117	91	3.0	20	0.11	21.2	12.5	3.0	1.9
9	120.58	28	0.23	0.12	104	87	3.3	21	0.15	21.3	10.7	3.1	2.0

Appendix. E. Heavy metal concentrations and yield of Puccinellia maritima (1984).

					BOO					-1		
Por nr.	Fresh weight			•	•							
	shoot	8	Pb	Pe Pe	¥	5	Zn	As	Na	×	క	₩
drained,	high selinity											
21	130.21	0.17	0.58	210	62	4.4	14	0.29	8.4	16.0	6.0	8.0
22	134.87	0.35	1.20	450	79	5.7	16	0.43	8.9	17.3		-
23	127.23	0.13	0.16	117	65	5.3	11	0.11	7.3	21.5	1.3	-
24	141.88	0.13	0.24	88	99	6.2	13	0.12	5.5	19.5	6.0	6.0
25	147.49	0.19	0.34	176	69	4.3	13	0.19	8.5	20.0],]	
drained,	low salinity					1) }) -))) •		;
5 6	141.02	0.25	1.32	829	11	3.5	15	0.57	6.3	20.6	۲,	C
27	127.32	0.39	2.54	1000	78	4.3	19	0.75	11.0	22.6	2.3	8
28	124.00	3.29	0.46	204	77	3.9	12	0.18	5.8	22.1	1.1	
29	155.77	0.28	99.0	259	54	3.7	12	0.15	6.6	16.6	1.1	6
30	137.00	0.29	0.30	115	71	2.8	10	0.12	8.4	17.6	1.0	9
inundated	d, high salinity					ı	;) -) !) •	•	
31	97.27	90.0	0.14	97	63	3.6	11	0.07	0.6	16.3	1.2	,
32	132.59	0.19	1.06	343	75	5.2	14	0.31	13.3	18.8	1.9	, R
33	109.13	0.10	0.54	126	54	3.8	12	0.14	16.0	17.0	1.6	2.6
34	124.62	0.09	0.86	425	99	4.0	10	0.39	11.3	38.3	1.4	1.6
35	104.78	0.13	96.0	306	92	4.5	10	0.24	20.8	8.84	4.1	2.1
inundated	d, low salinity))))	,	•
36	122.93	0.24	09.0	230	99	2.9	11	0.21	4.8	15.6	1.3	45
37	116.24	0.05	0.34	131	26	2.6	10	0.18	8.5	18.1	1.2	
38	175.10	0.19	0.52	149	57	2.7	11	0.20	0.6	13.6		7
39	158.79	90.0	0.26	226	65	4.2	0	0.14	6.6	15.8	1.3	
40	127.23	0.03	0.20	104	65	3.1	6	0.07	7.9	14.2	1.0	1.1
												† •

Appendix. F. Heavy metal concentrations and yield of Aster trivolium leaves (1984).

					BOO					18 · 8		
Pot nr.	Fresh weight shoot	8	Pb		W	ಕ	Zn2	As	N.	×	క	<u>\$</u>
drained,	high salinity			!	•	,	:	Ġ	ć	Š	6	4
-	144.84	0.59	0.54	95	159	7.7	46	0.04	12.3	20.0	7.0	7
7	168.03	0.78	0.76	9	120	5.3	45	0.01	46.4	24.8	4.1	2.0
· (~	0.0	ı	i	ı	1	1	ı	,	ı	ı	ı	1
•	156.18	0.51	0.84	153	144	7.5	27	0.05	65.0	28.3	6.2	2.3
'n	96.67	0.44	0.42	19	120	6.6	30	0.02	76.0	29.0	5.2	æ.
drained.	2										,	•
9	51.68	0.83	0.48	118	181	٠٤ وي	98	0.15	53.0	25.1	9.9	7.1
^	161-13	0.52	0.36	72	87	5.2	31	0.04	45.2	26.1	5.5	1.8
. cc	156.93	0.59	09*	89	20	9.9	53	0.04	43.7	44.4	5.1	2.2
) o	57.36	3.14	0.70	72	281	5.1	138	90.0	52.5	24.6	11.2	2.4
01	61.00	2.33	0.70	69	225	6.7	103	90.€	49.2	56.6	9.6	2.2
inundated,	d, high salinity							•	•	•	,	
11		0.17	0.54	55	81	5.5	33	0.04	58.8	23.8	4. 9	1.3
12	71.79	0.37	0.72	29	70	3.6	54	0.10	45.0	35.5	3.6	4.6
13	93.18	0.24	0.52	81	106	5.4	32	0.12	60.5	25.5	5.9	1.7
14	154.00	0.17	0.32	96	102	5.8	25	0.09	63.6	27.0	4.8	1.4
15	105.28	0.16	0.50	78	9/	2.6	31	0.13	72.4	32.4	5.4	1.6
foundated,	d, low salinity								•	,	•	•
16	69.06	0.26	0.52	180	163	2.6	45	0.11	58.0	14.8	4.4	
17	157.82	0.18	0.52	58	9	2.6	18	0.01	65.0	25.1	9.9	2.1
· C	218.58	0.36	0.80	48	112	8.9	22	0.05	80.6	23.3	8.1	2.5
6	212.38	0.19	0.44	57	71	3.4	22	0.03	54.2	26.9	5.1	1.5
5 0	15.26	2.04	1.76	375	294	5.5	120	0.36	87.9	22.6	14.2	3.6

Appendix. G. Heavy metal concentrations and yield of Aster tripolism stems (1984).

					E00					7		
Pot nr.	Fresh weight		·							8.8		
	ates:	ਝ	Pb	Fe	¥	Ş	Zn	As	Na S	×	త	¥
drained,	high salinity				٠							
-1	103.68	0.27	0.12	152	31	7.6	17	0.13	17.6	13.9	1.4	0
7	27.12	0.43	0.46	124	21	16.3	27	0.05	4.6	4.3	7	
m	6.53	1.10	2.38	842	75	29.5	89	1	54.5	23.0	1 4	
•	18.24	0.54	1.00	477	36	19.0	23	0.33	7.5	0.4	8	<u></u>
5 0	23.19	67.0	1.02	434	38	22.9	35	97.0	10.8	0) <
drained,	low salinity					i 				?		•
•	79.77	0.63	09.0	146	35	4.7	25	0.20	32.4	17.6	2.6	1,0
_	8.99	0.59	0.70	96	20	17.4	21	0.12	6.3	6.3	2.1	7 -
€	3.68	0.59	0.70	96	20	17.4	21	0.12	6.9	6.4	2,1	
Φ.	52.24	1.57	0.14	50	42	6.7	30	0.08	15.6	15.1		
01	48.76	1.14	0.20	42	37	8.2	28	0.07	18.1	1.01	7 4) ·
Inundated	i, high salinity				,		ì		•	1.01	•	7.1
11	21.36	0-0	0.22	146	14	10.9	16	0.12	8.8	7.8	7.4	7
12	5.24	C.68	4.06	1561	99	14.6	31	•	11.0	8.9	3.0	2.1
13	18.78	0.16	0.86	149	16	14.5	21	0.21	8.0	4.0	1.5	
14	21.53	0.14	0.83	397	21	13.2	18	0.35	6.4	8.9	1.6	7
15	17.79	0.23	0.80	365	29	13.0	31	0.51	16.5	S . S	,	
Inundated	1, low salinity						1	1		•	•	
16	85.90	0.18	3.48	218	62	5.1	21	0.11	31.6	12.6	7 5	
17	14.71	0.17	0.64	301	21	13.4	17	9.0	13.6	0.4	4.0	•
18	23.37	0.22	050	142	23	12.8	19)	16.3	. 4		•
19	18.55	0.16	032	120	17	11.8	15	0.13	8	7	, , ,	7
20	38.14	0.41	95.0	148	33	7.8	31	0.14	30.1	15.6) a	7 -
)		•	•	•	7

Appendix H.

	K(X)
sambres/	Ca (X)
(plant	ಕ
Quality control (plant samples,	8
Quality	£
	į
	.

	ģ	5	£	ខ	£	8	ಕ	Ca(X)	K(Z)	Mg(1)
	U L	į	ļ		•		C	2.50	4.56	0.68
	752	64	237	5.6	6.0	J• C	ָר בּי	2.93	4.21	0.63
	705	57	228	9.3	٠,٠	י י	0.4	3.11	4.58	89.0
	700	09	244	6.6	0.0	, ,	6.4	3.03	4.30	99.0
	739	62	238	10.3	o «		2.5	2.95	4.46	99.0
	722	61	239	4.6	7.0		0	3.00	4.51	0.68
	745	59 61	244 244	e. e.	. e. e	. e.	2.0	3.06	4.46	0.68
	6	Ş	910			3.4	5.0	3.00		0.67
Estandard deviation	22.2	2.22	5.78	0.351		0.177	0.105	0.028	0.052	0.0071
standard error	8.40	0.84	61.7				•	2.5		2.8
coefficient or variation	3.1	3.7	2.4			5.2 0.15	0.10	0.07		0.01
95% confidence	70	7	n			ò	a c	0.03		0.01
limits of detection limit *	0.45	0.05	0.04 238±7		2.2 6.3±0.3	3.6	4.5±0.5 3	3.00±0,03		03 (0.7)
NBS 1573 (certified values)	674060	}				بر ا	to the state of th	.		

* standard deviation x 3 (n=6) Fe, Zn, Hn, Ca, K, Hg in ppm en Cu, pb, Cd, Cr in ppb () = non-certified values, information only

Samples are analysed according to the analysis methods of the soil acience laboratory DIHO, 1984.

15 February 1985 - Soil Science Laboratory